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A REVIEW ON GREEN SYNTHESIZED NANOPARTICLES AND ITS IMPORTANCE IN CANCER THERAPEUTICS

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ABSTRACT

Nanoparticles play an important role in Science and Technology. As, nanoparticles are nano in size they have their unique properties due to its surface volume ratio. This review article, highlights the importance of nanoparticles in cancer therapeutics and its applications in drug delivery and its biocompatible nature.

KEYWORDS: Nanoparticles, Science, Technology, Drug delivery, Biocompatible.

INTRODUCTION

The term nanotechnology was first proposed by physicist Richard Feynman in 1959 in his talk entitled "There's Plenty of Room at the Bottom" presented at the American Physical Society meeting at the California Institute of Technology in 1959. The term nanotechnology was defined by Japanese scientist Dr. Nori Taniguchi in 1974 as "the processing of separation, consolidation and deformation of materials by one atom or one molecule".[1,2] Nanoscience, nanotechnology, nanomaterials, nanooncology are some of the terms containing nano. The prefix "nano" came from the Latin word "nanus" literally means dwarf very small. Nanotechnology has rationalized the world of science and technology which deals with small things that are less than 100nm in size.[3] Nanomaterials have significantly different properties than the same materials at larger scale. Nano sized particles have endless possibility due to their unique properties.[4]

Among all the nanoparticles metallic nanoparticles have been used in drug delivery systems especially for the treatment of cancer and biosensors.[5] In recent years, an exponential interest has developed for delivering novel drugs using nanoparticles.[6] Nanoparticles can be used in different route of drug administration due to its high stability, high specificity, high drug carrying capacity, controlled release of drugs and the ability to deliver both hydrophilic and hydrophobic drug molecules in the diagnosis and treatment of diseases.[7] Bio-nano interactions showed the relations between nanoscale entities and biological systems such as peptides, proteins, lipids, DNA and cellular receptors.[8]

In terms of global nanoscience India was ranked 4th position, possessing about 500 nanotechnology companies and 200 nano products commercially available in the market.[9]

Nanoparticles as nanocarriers

Cancer is the second leading cause of death next to cardiovascular disease. The current clinical treatments are unable to provide timely detection and curative therapy. To overcome these limitations nano oncology field plays a vital role. This field have designed new strategies to deliver chemotherapeutic drugs to the tumor site at higher concentrations with minimal damage to normal tissues.[10]

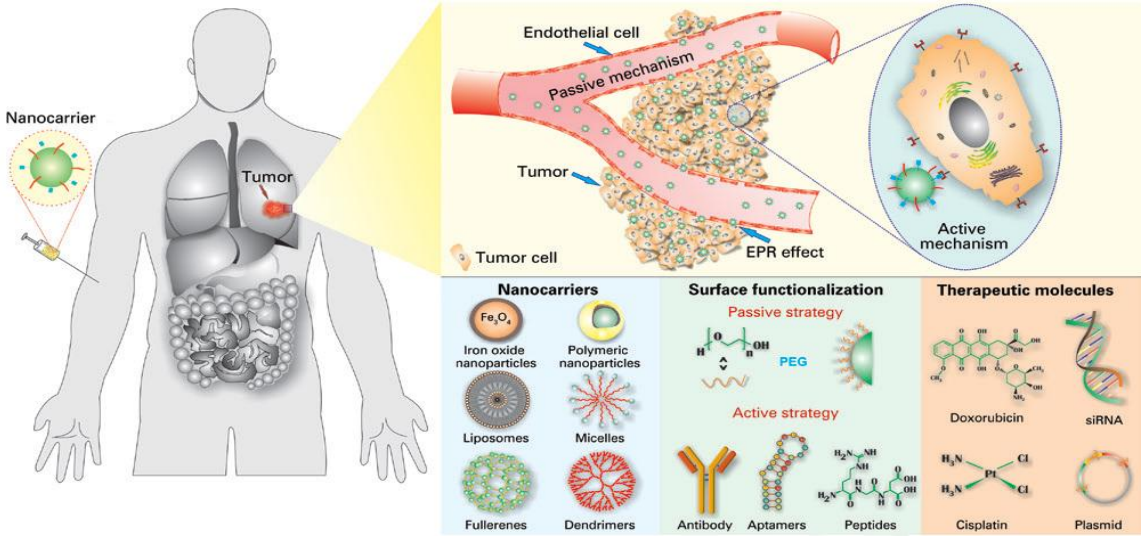

Figure 1: Nanocarriers for cancer treatment.

Green synthesized nanoparticles from medicinal plants

Green synthesis of nanoparticles using renewable natural sources like plant extract, microorganisms and biodegradable sources which acts as a reducing and capping agent have drawn tremendous attention towards nano field. For the rapid synthesis of nanoparticles plant sources are widely used as they fit for large scale biosynthesis. Various plant parts such as

seed, leaf, stem, root and latex are commonly used for metallic nanoparticles synthesis [11]. Employing green technology for the synthesis of nanoparticles was found to be the best method, as it does not involve any harmful chemicals.[12] Moreover, the synthesis of nanoparticle was influenced by the nature of the plant extract, pH, temperature, concentration of the metal salt and the contact time.[13] It has been proven that, plant derived silver nanoparticles acted as a good antioxidant agent as silver capped with the active functional groups present in the phyto constituent of the plant extract.[14]

Biomedical potential of green synthesized nanoparticles

Magnetic nanoparticles have gained vast interest in the field of biomedicine due to its wide applications such as molecular imaging, photothermal therapy, magnetic hyperthermia and as drug delivery vehicles. The multimode nature of these nanoparticles can act as both therapeutic and diagnostic tools (theranostic agents).[15] Silver nanoparticles are used commonly for biomedical applications such as antimicrobial coatings, wound dressings and biomedical devices.[16]

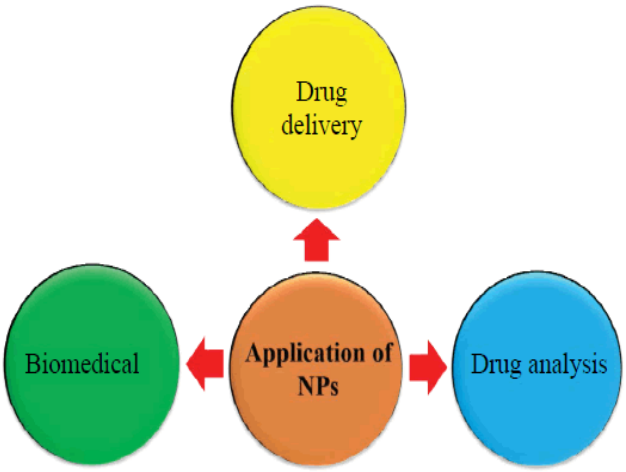

Figure 2: Applications of nanoparticles.

Biocompatibilty of nanoparticles

Biocompatibility first drew the attention of researchers between 1940 and 1980 in the context of medical implants and their interaction, both harmful and beneficial effects within the body. Recently the concept of biocompatibility was more pronounced and formally defined as “The ability of a material to perform with an appropriate host response in a specific situation”. The three dogma which play important role are that a material has to perform its intended functions and not merely be present in the tissue, that the induced reaction has to be proper

for the intended application, and that the nature of the reaction to a particular material and its suitability may be different from one context to another.[17]

In 2010, Kohane and Langer.[18] explained biocompatibility in the context of drug delivery and defined biocompatibility as "an expression of the benignity of therelation between a material and its biological environment". However, some researchers have expanded the definition of biocompatibilty by denoting acceptable functionality of a biomaterial. High degree of biocompatibility is achieved when a material interacts with the body without inducing unacceptable toxic, immunogenic, thrombogenic, and carcinogenic responses.[19]

When nanoparticles interact with the body, a variety of responses may occur. These include alterations in the immune system or interaction with blood. These reactions vary significantly with nanoparticle composition.[20] For example, gold nanostructures may interact differently with the body when compared to polymeric particles. For this reason, nanoparticles have to be evaluated individually or "on a case-by-case basis" to understand their effect on the body in a better way.[21]

To establish a safe nanotechnology it was necessary to study the nongenotoxic nature of the nanomaterials. Several genotoxicity assays can be carried out *in vitro*. *In vitro* cytotoxicity studies of nanoparticles using difference cell lines, with different nanomaterials are increasingly studied published. The techniques that can be used to assess toxicity of nanomaterials include:

1) *In vitro assays* for cell viability/proliferation using MTT, LDH assay and mechanistic assays [ROS generation, apoptosis, necrosis and DNA damaging potential] using ROS assay
2) Microscopic evaluation of intracellular localization using SEM-EDS, TEM
3) Gene expression analysis, high-throughput systems
4) *In vitro* hemolysis and
5) Genotoxicity

The biocompatibility study of nanoparticles were carried out in pulmonary, erythrocytes and endothelial cells for cardiovascular disease, ovarian cancer, in animals spleen injury, lung inflammation, mouse embryonic fibroblasts, human monocytes, human spermatozoa, murine glioma cells and ocular use.[22,23]

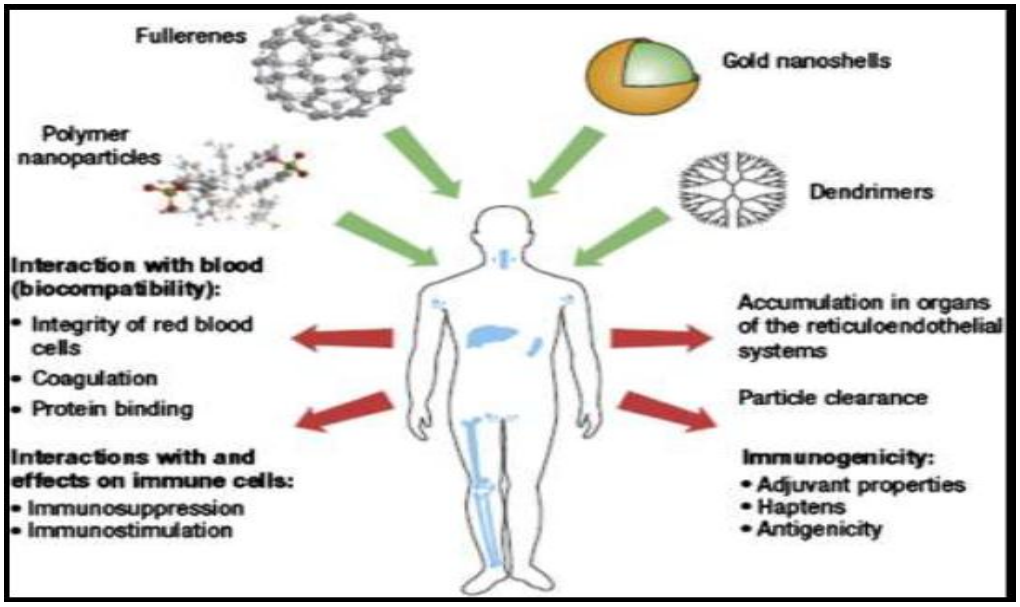

Figure 3: Biocompatibility of nanoparticles.

Drug releasing profile of nanoparticles

Nanotechnology has been utilized in medical field for therapeutic drug delivery system and the development of treatments for a variety of diseases and disorders. So, there are very significant advances in these disciplines. In the early 1970s, controlled drug delivery systems aimed to deliver drugs at predetermined rates and predefined periods of time, have drawn increasing attention.[24]

On the other hand, drug delivery is an emerging field focused on targeting drugs or genes to a desirable group of cells. The ultimate goal of this targeted delivery is to transport proper amounts of drugs to the desirable sites such as tumors, diseased tissues etc. while minimizing unwanted side effects of the drugs on other tissues.[25]

Recently numerous proteomic and drug design profile have emerged to target various cellular processes, creating a demand for the development of intelligent drug delivery systems that can respond directly to pathophysiological conditions. Micro and nano scale intelligent systems can maximize the efficacy of therapeutic treatments in numerous ways because they have the ability to rapidly detect and respond to disease states directly at the target site, sparing physiologically healthy cells and tissues and thereby improving a patient's quality of life. This new class are designed to perform various functions like detection and release of therapeutic agents for the treatment of diseased conditions. Stimuli responsive biomaterials are very promising carriers for the development of advanced intelligent therapeutics.[26]

Polymeric NPs are composed of biodegradable/ biostable polymers and copolymers. The drug molecules can be entrapped or encapsulated within the particle, physically adsorbed on the surface or chemically linked to the surface of the particle.[27]

Nanoparticles as free radical scavengers

Rapid synthesis of metallic nanoparticles with appropriate morphology and sizes have drawn the attention of researchers due to its evocative therapeutic applications such as antioxidant, antibacterial, anticancer, larvicidal, catalytic and wound healing activities. The *in vitro* antioxidant activity of the nanoparticles was determined by their efficiency to scavenge free radicals. The silver nanoparticles produced from the aqueous extract of *A. Marschalliana showed a potent in vitro antioxidant activity by scavenging DPPH* 2, 2-diphenyl-1-picryl hydrazyl assay.[28] Nagaich *et al.* (2016).[29] reported that the hydrogels loaded silver nanoparticles synthesized from the flavonoids of apple extract exhibited high radical scavenging activity by DPPH assay with the per cent radical inhibition of 75.16% ± 0.04. The gold and silver nanoparticles synthesized from the aqueous extract of *Solanum torvum* (*S. torvum*) fruit served as strong radical quenching by effectively scavenging hydroxyl, superoxide, nitric oxide and DPPH radical.[30]

Nanoparticles as antioxidants

Antioxidants are substances that inhibit oxidative damage by scavenging free radicals and protect the human body. Imbalance between antioxidants and free radicals leads to oxidative stress which results in cellular damage.[31] Our body naturally produces free radicals as a by-product when cells utilize oxygen, the produced free radicals has the tendency to cause damage. Antioxidants has the capacity to scavenge free radicals and protects the humans against heart disease, muscular degeneration, diabetes mellitus, cancer and aging are all contributed by oxidative damage. Nanoparticles as antioxidants counteract the oxygen levels and ROS production.[32] Sriramulu and Sumathi (2017).[33] reported that the silver nanoparticles synthesized from mushroom explored potent *in vitro* antioxidant, anti-inflammatory and antimicrobial activity against *Escherichia coli* and *Staphylococcus aureus*. Direct delivery of antioxidant enzymes to cells is highly impossible. To overcome these difficulties design of nanoparticles to deliver enzymes is important for therapeutic purposes. Lin *et al*., 2016.[34] resulted that mesoporus silica nanoparticles fused with TAT peptide simultaneously delivered two upstream antioxidant enzymes SOD and GPx which exerted synergistic effect to cell by scavenging ROS.

Nanoparticles as anti lipid peroxidative agent

Lipid peroxidation (LPO) is an autocatalytic process causes cell death. The reaction is initiated by the decomposition of lipid peroxides which produce reactive carbonyl compounds. The by-products of lipid peroxidation are MDA and LH involved in cataractogenesis, mainly due to its cross linking ability. A free radical attack on cell membrane PUFA initiates the autocatalytic process which generates large amount of toxic radical products which initiates tumor initiation and promotion of colorectal cancer.[35] Green synthesized phyto nanoparticles acts as a good antilipid peroxidative agent. Pathak *et al*., 2015.[36] reported that nanocarriers formed by the assembly of two oppositively charged lipid and polymer for curcumin acted as an antilipid peroxidative agent determinted by *in vitro* TBARS assay the encapsulated had a great potential as functional food ingredient than the un encapsulated form.

Nanoparticles as apoptotic agents

Nanomedicine has revolutionized this modern era, by designing personalized drugs for cancer treatments by providing a clear-cut solution faced by the current systemic therapeutic drugs. Combinations of drugs and nanoparticles increases the synergetic approach which provides greater therapeutic effects than does single drug treatment.[37] Nanoparticles combined with different anticancer drugs is an advanced therapeutic strategy. Nanoparticle mediated targeted drug delivery systems permits the drug to accumulate in tumors, by either active or passive targeting and increases the intracellular drug concentration and substantially enhances the cytotoxic effect of various antitumor agents.[38] Plant kingdom produces a vast naturally occurring secondary metabolites, nano-medicine enhances the anticancer activities of the plant derived drugs by converting them to nano form. Nanoparticles synthesized by plant mediated source typically enhances the apoptotic activity by its targeted controlled release of the drug.[39] The biosynthesized silver nanoparticles provides a better therapeutic frame work as apoptotic agent against a cluster of cells in the initial stages of cancer by overwhelming the biological barriers.[40] Biogenic silver nanoparticles became a potential cancer therapeutic, diagnostic and apoptotic agent in the current era.[41]

Nanoparticles in DNA fragmentation

Apoptotic cell death is characterized by certain morphological and biochemical changes which distinguish from other forms of cell death. DNA fragmentation is an important feature

of apoptosis.[42] Caspase-Activated DNase (CAD) is the enzyme responsible for DNA fragmentation. This enzyme cleaves DNA at inter nucleosomal linker sites between nucleosomes. Degradation of nuclear DNA into nucleosomal units and formation of DNA fragments is one of the hallmarks of apoptotic cell death.[43] A conventional agarose gel electrophoresis was commonly used to analyze fragmented nuclei in cell.[44] Biogenic silver nanoparticles showed effective cytotoxicity and antiproliferative effect than chemically synthesized silver nanoparticles. The induction apoptotis by DNA fragmentation was found to be highly effective in the synthesized silver nanoparticles against K562 leukemia cell line.[45]

Nanoparticles in cell cycle regulation

Cell cycle analysis was studied by flow cytometry measurement. Flow cytometry is a laser based instrument used in cell counting, cell sorting, biomarker detection and protein engineering. In flow cytometer the cells are suspended in a stream of fluid which passes through an electronic detection apparatus.[46] There are four widely used methods to analyze cell cycle by flow cytometry. The first two methods are based on analysis of cellular DNA content followed by staining the cells with either propidium iodide (PI) or 4′, 6′-diamidino-2-phenylindole (DAPI) based on univariate analysis. These methods expose the distribution of cells in three phases of the cell cycle (G_1 vs S vs G_2/M) which enables to detect apoptotic cells with fractional DNA content. The third method is based on analysis of DNA content and proteins associated with proliferation. This method enables to distinguish cells, in different phases of cell cycle identify mitotic cells or intracellular expression of proteins to the cell cycle position. The fourth method depends on the detection of 5′-bromo-2′-deoxyuridine (BrdU) labelled on the DNA replicating cells.[47]

Nanoparticles as antitumorigenic agents

An important challenge in the field of Oncology was to find a technology for the controlled targeted release of the drug and eradicate tumor cells by sparing normal cells.[48] In this context, biodegradable nanometer sized particles gained great interests among pharmaceuticals due to its novel structural and physical properties for targeted delivery of anticancer agents and imaging contrast agents.[49] Nanocarriers are used to overcome the side effects associated with conventional antitumor drugs such as their non-specificity, burst release of the drug and damaging the normal cells. Moreover, nanocarriers improve the bioavailability and increase the therapeutic efficiency of the

antitumor drugs at the target site by preferential accumulation.[50] Nanoparticles synthesized from medicinal plants targeting biological pathways has tremendous advantages due to its higher efficacy and fewer side effects compared to commercial cancer drugs.[51,52]

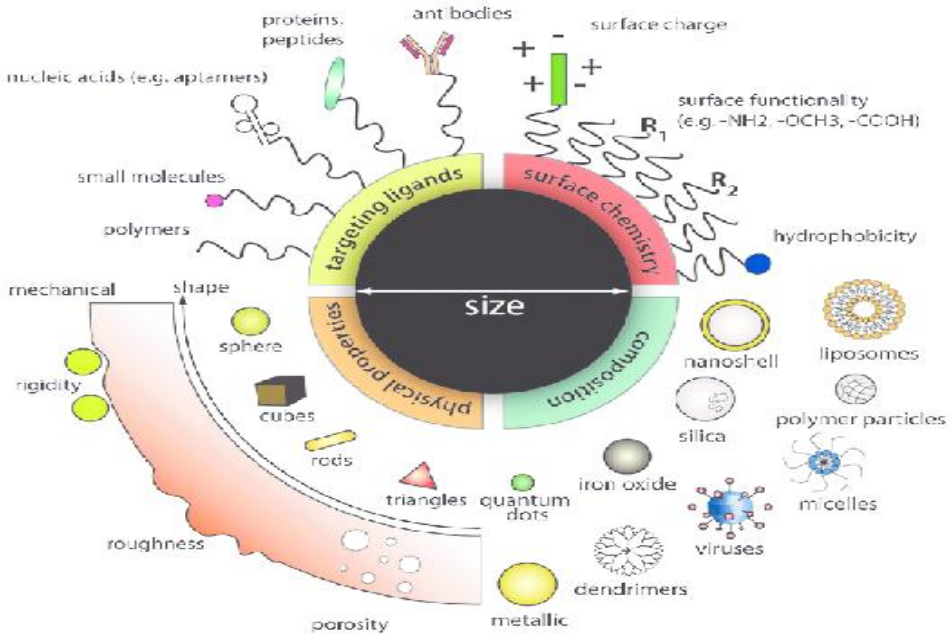

Figure 4: Design of nanoparticle based drug delivery platform.

CONCLUSION

Green synthesized nanoparticles have gained advantage, as these nanoparticles are less toxic and eco-friendly. To conclude, Plant derived bioactive constituents are one of the active bio resources for the green synthesis of nanoparticles which enhances the antioxidative and antitumorigenic potential.

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